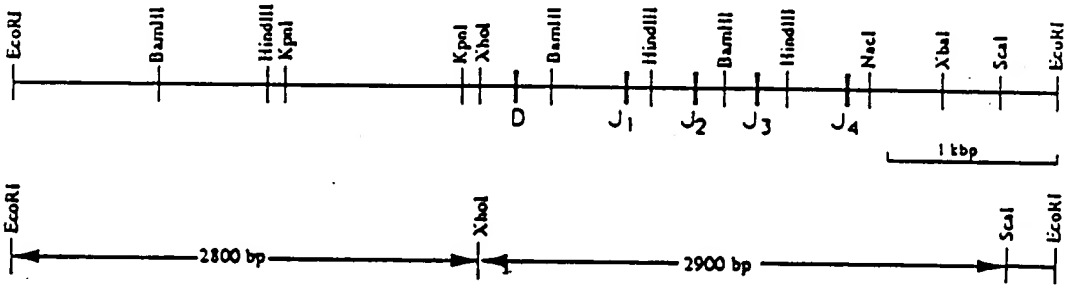
Mouse Heavy Chain J Genes Inactivation Vector

(A) Targeted mouse heavy chain J genes




(B) Inactivation vector mDAJ.Neo

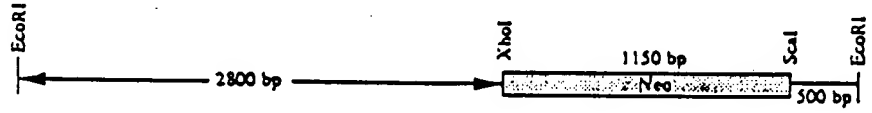
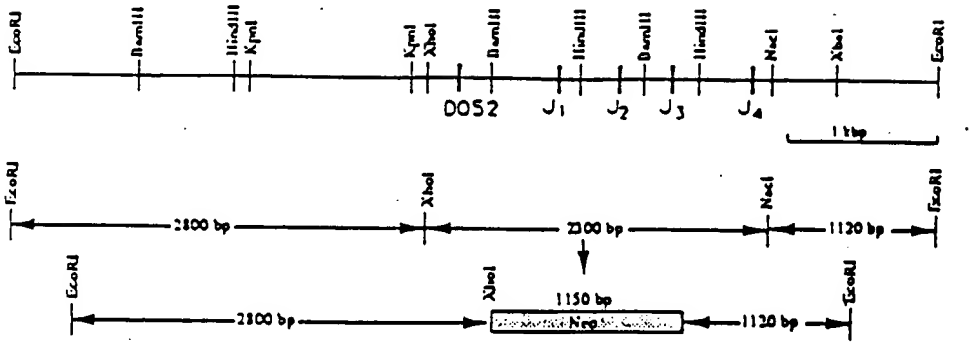


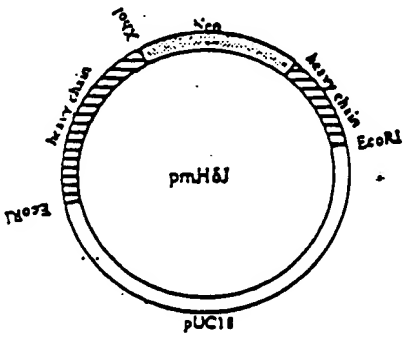

Figure 1

EI124 990720US

(A) Targeted mouse heavy chain J genes

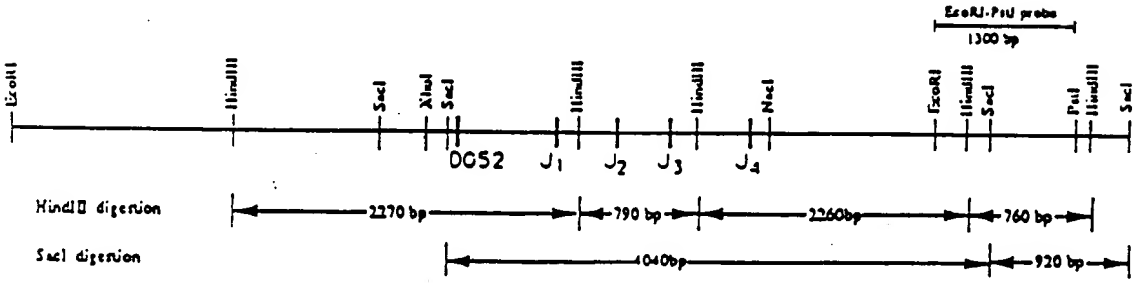



(B) Inactivation vector pmHδJ




(C) Southern analysis of pmHδJ-targeted ES colonies

Wild type ES cell genome




Targeted ES cell genome

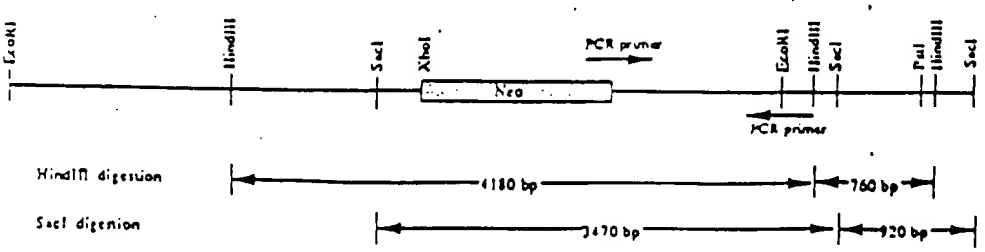
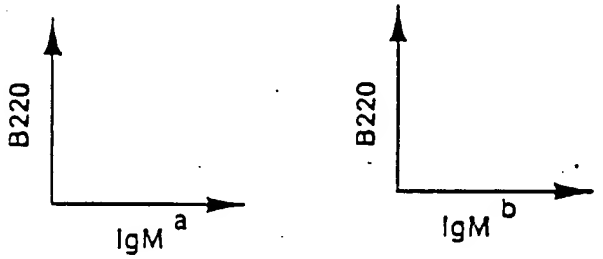
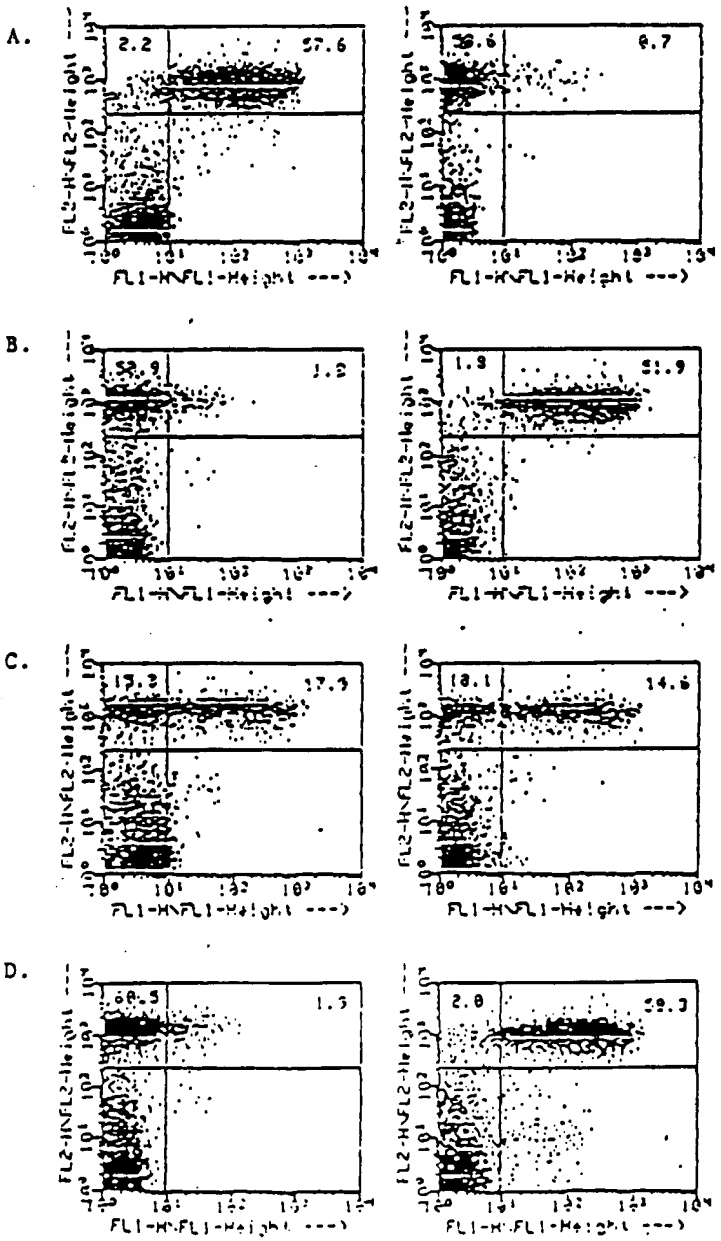



Figure 2

$J_H$ deletion blocks cell surface IgM expression




a allotype

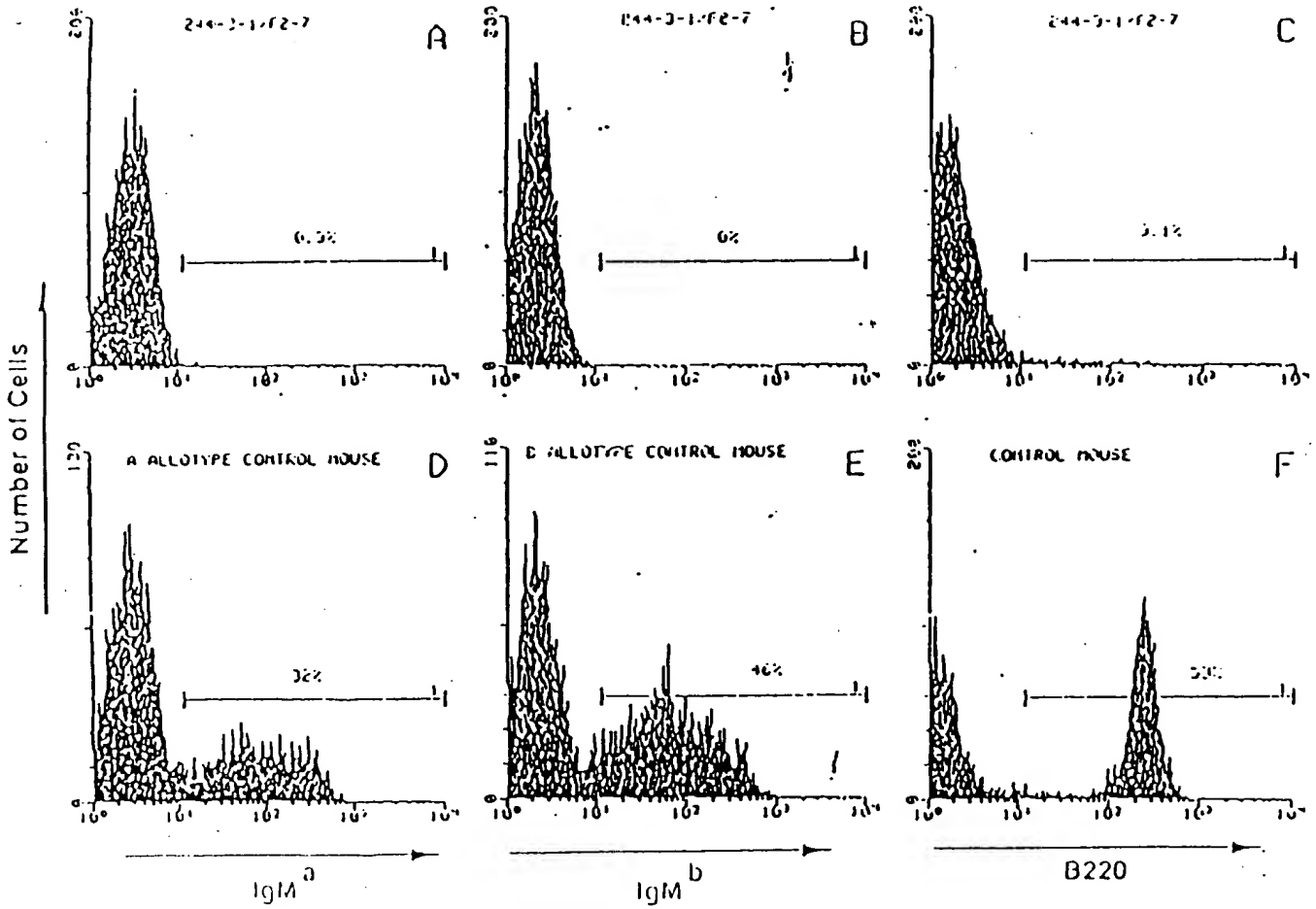
b allotype

a / b F1

$\Delta J_H$ / b F1

Figure 4

Staining of peripheral blood lymphocytes with fluorescent anti-a allotype (A, D), anti-b allotype (B,E) or anti-B220 (C, F). (A, B, C) JH-deletion homozygous mutant mouse 244-3-2/F2-7, (D) A allotype control mouse, (E) B allotype control (F) control mouse. The number in each panel indicates the percentage of cells stained with the specific antibody.



# INACTIVATION OF KAPPA CONSTANT REGION

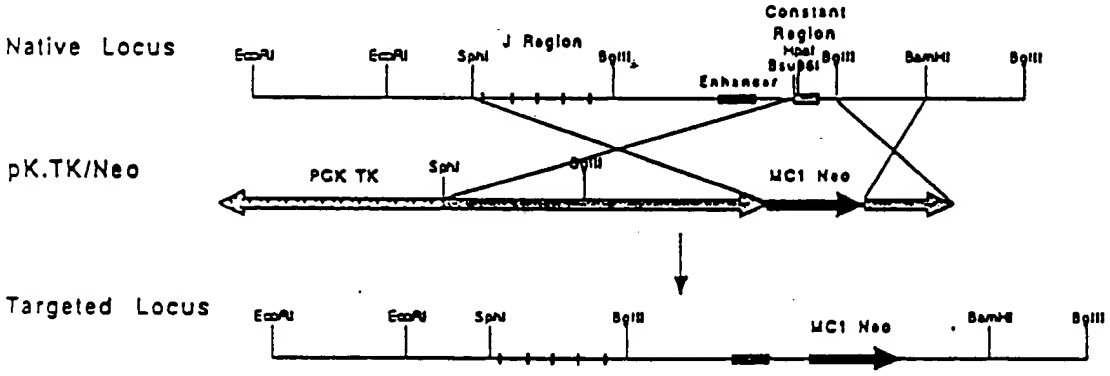



Figure 5

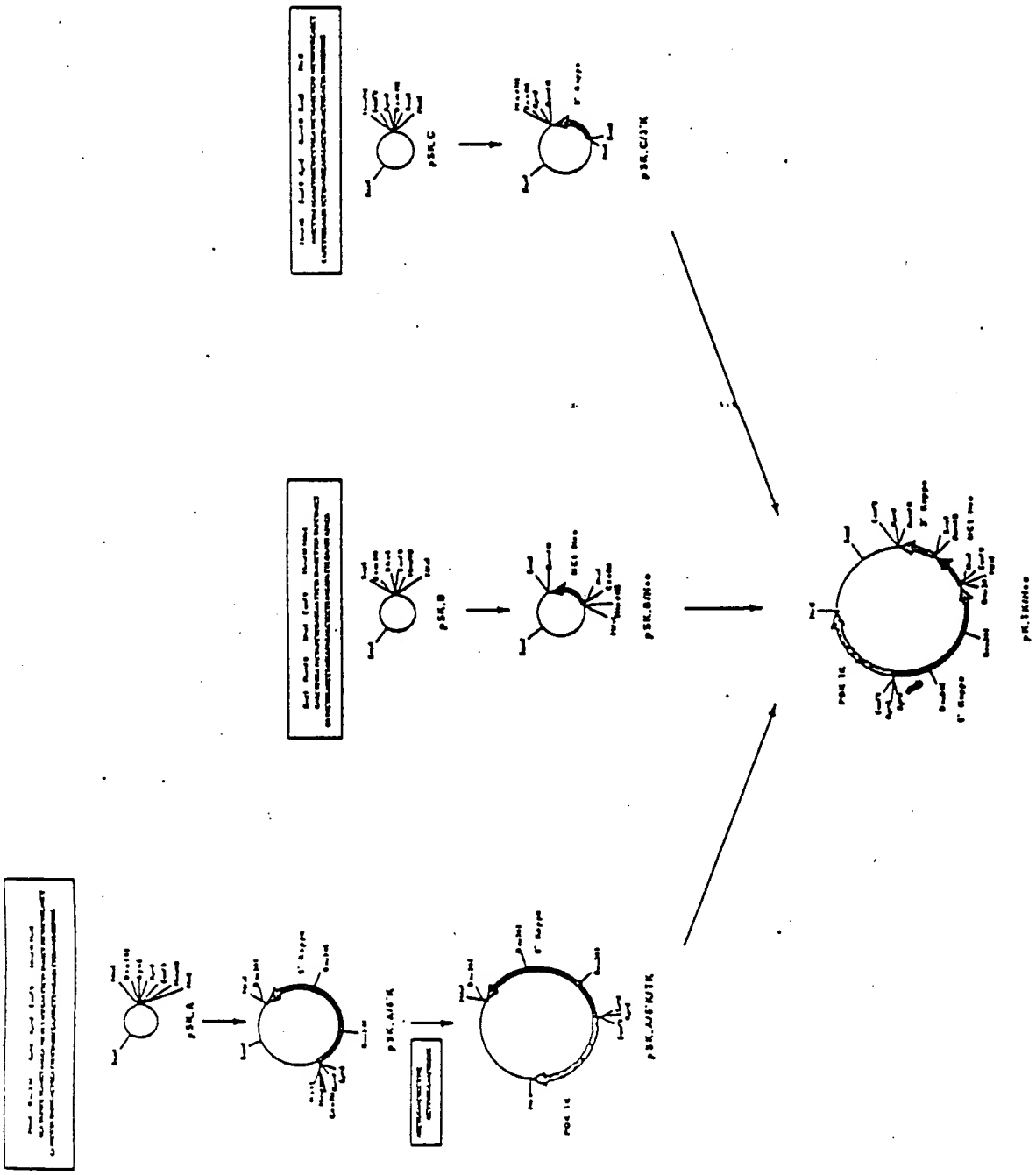
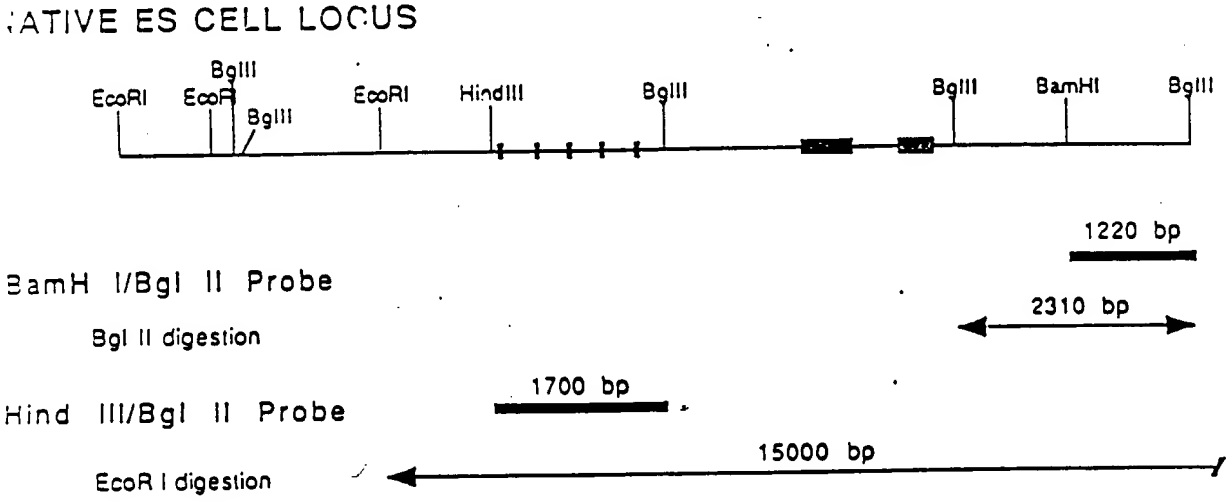


Figure 6

# SOUTHERN ANALYSIS OF LIGHT CHAIN Cκ-TARGETED E14-1 CELLS



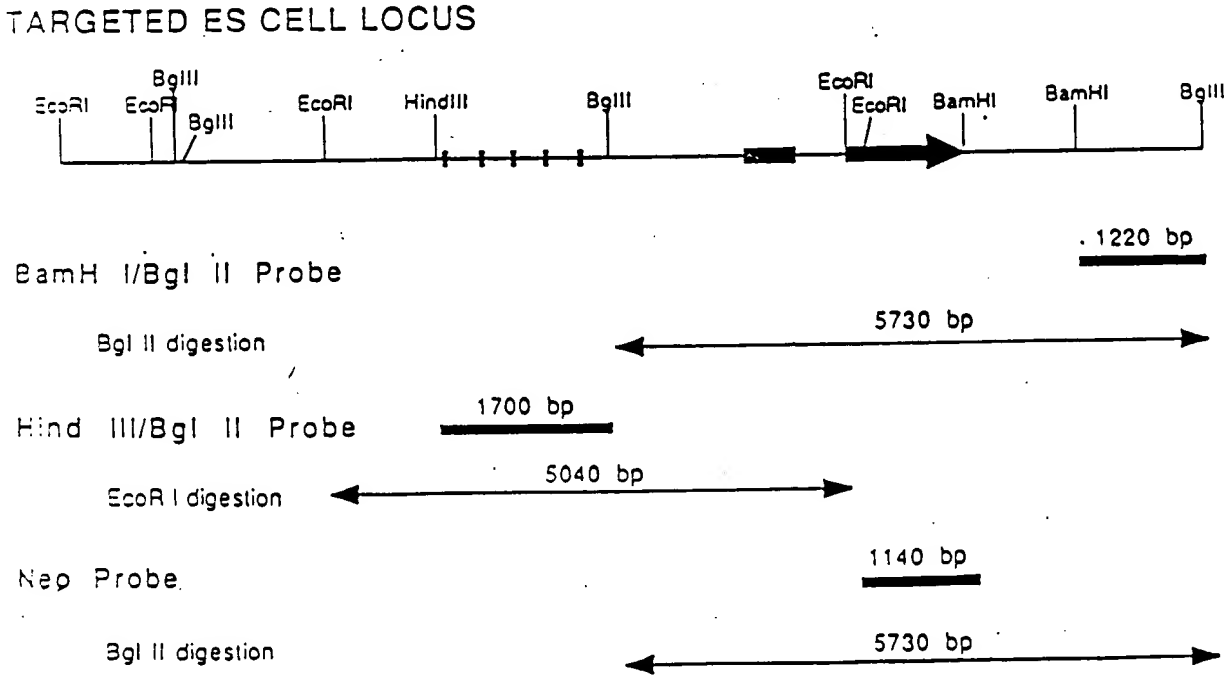
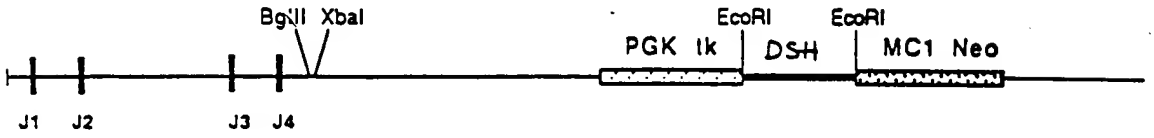




Figure 7

# KAPPA J/CONSTANT REGION INACTIVATION

## J REGION KNOCKOUT VECTOR




## TARGETING SCHEME

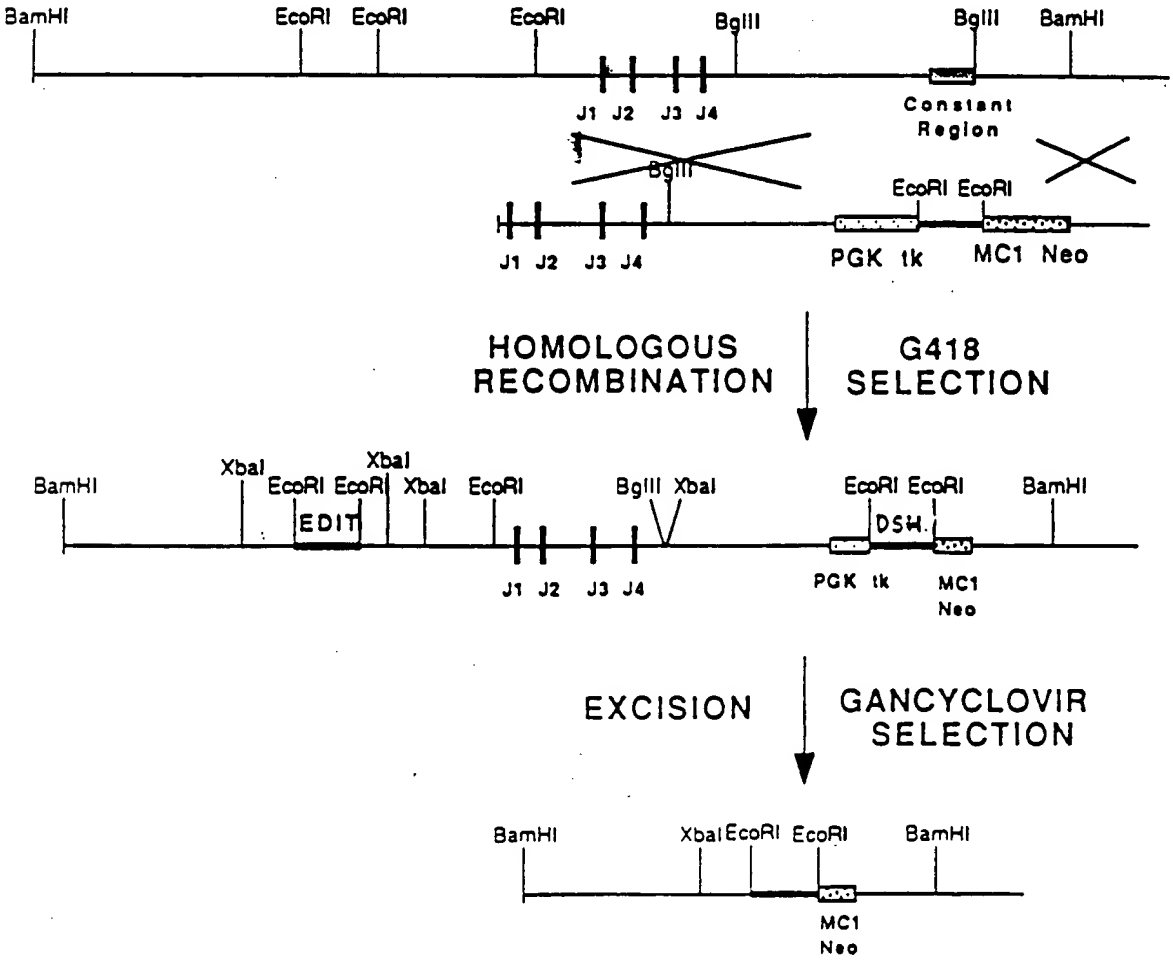



Figure 8

# CONSTRUCTION OF KAPPA J/CONSTANT REGION DELETION VECTORS

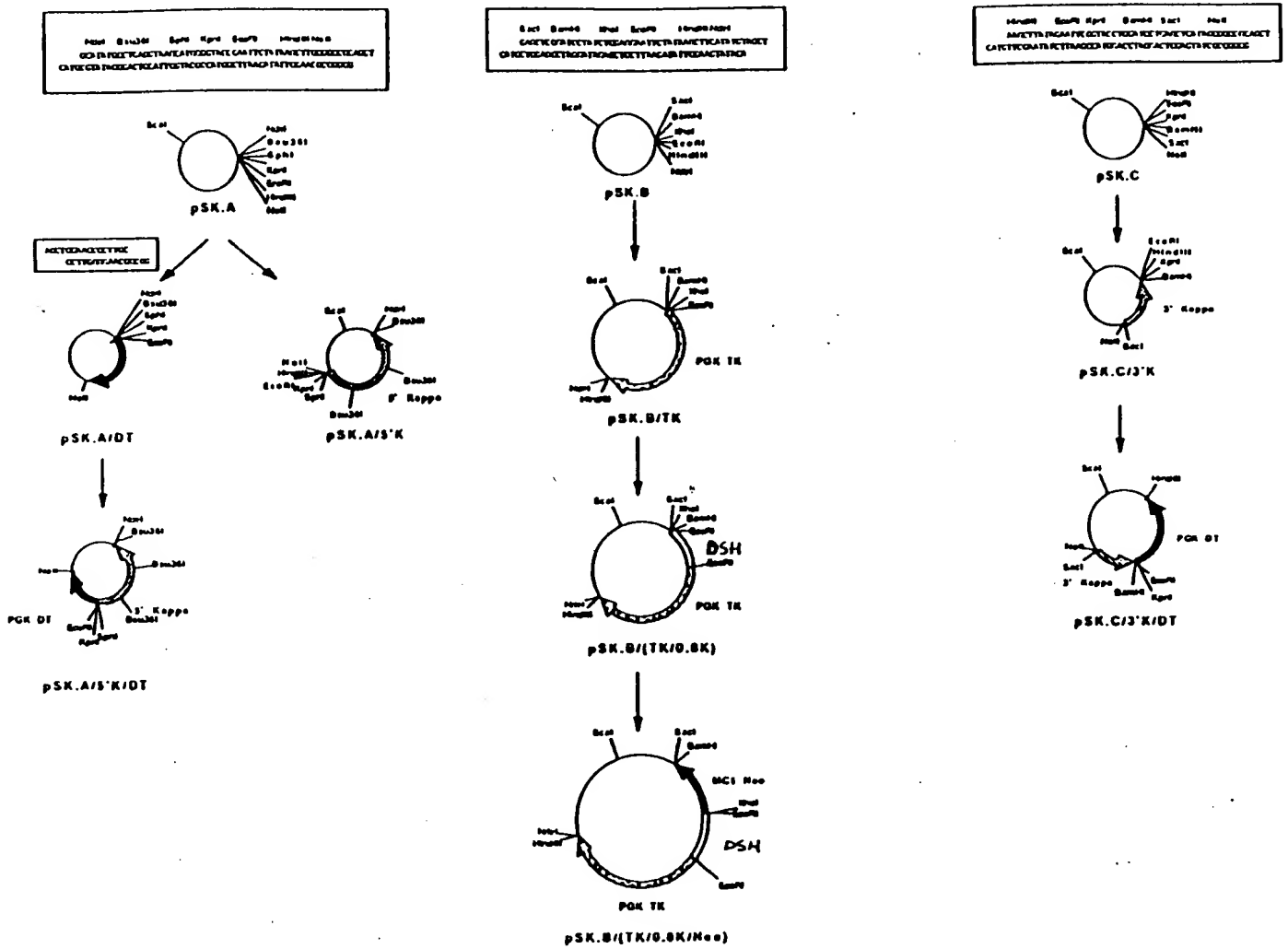
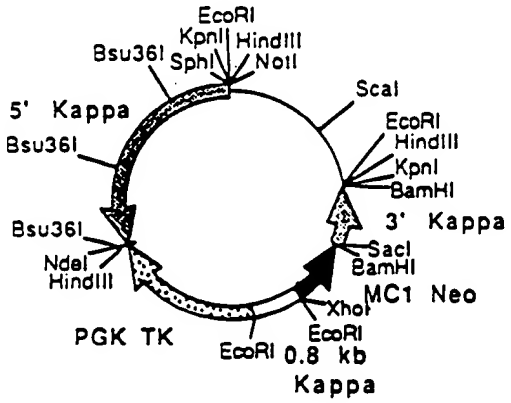
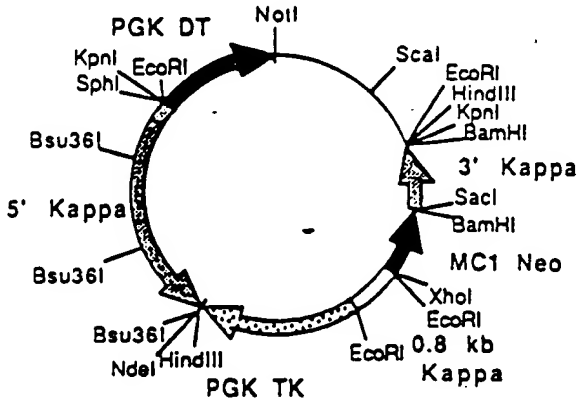



Figure 9

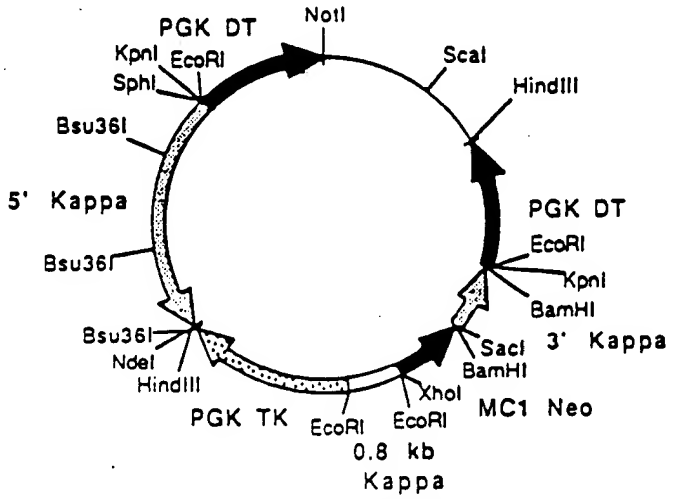
# KAPPA J/CONSTANT REGION DELETION VECTORS




pK.(TK/0.8K/Neo)




pK.DT/(TK/0.8K/Neo)




pK.DT/(TK/0.8K/Neo)/DT

Figure 10

# SOUTHERN ANALYSIS OF LIGHT CHAIN Jκ/Cκ-DELETED E14-1 CELLS

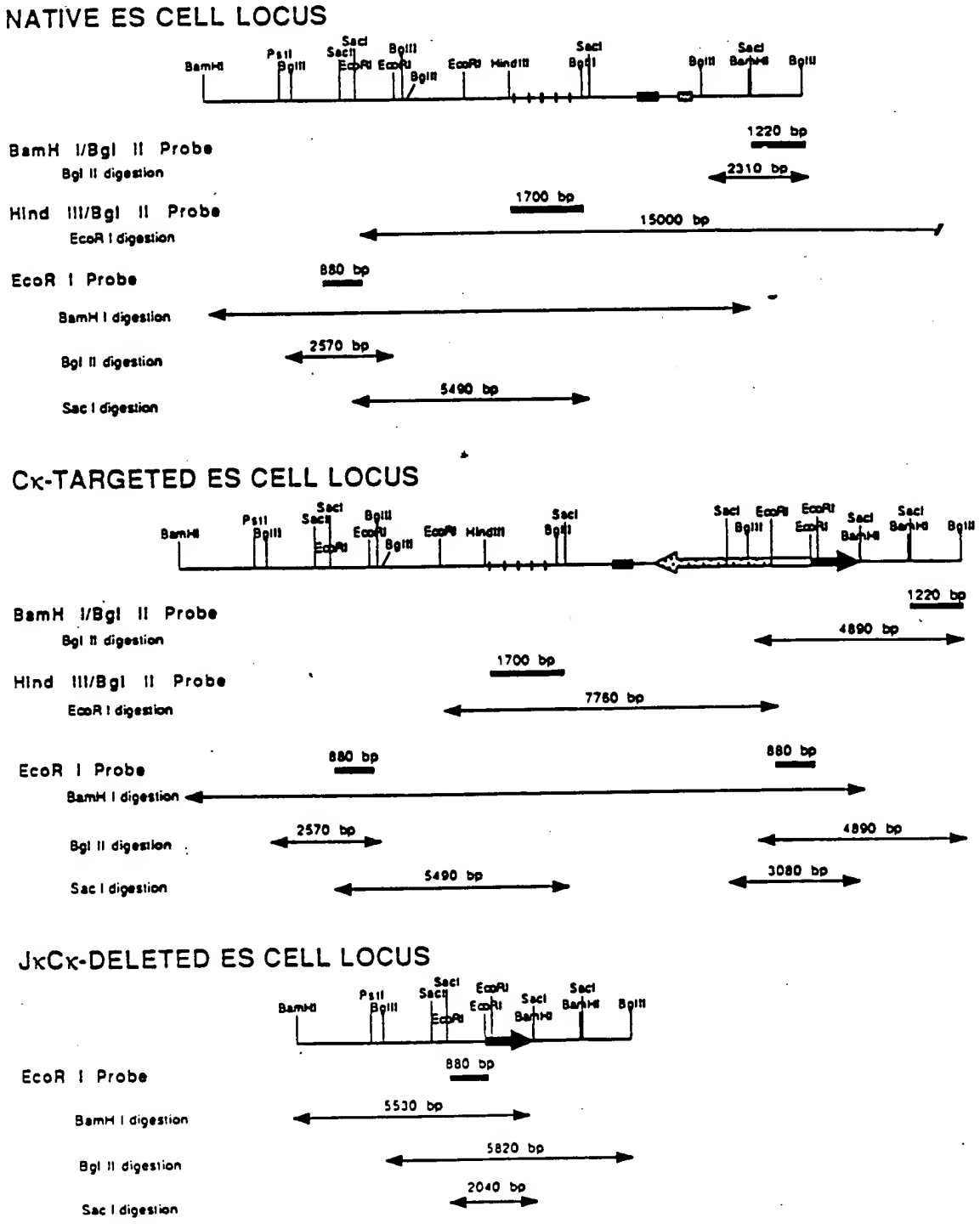



Figure 11

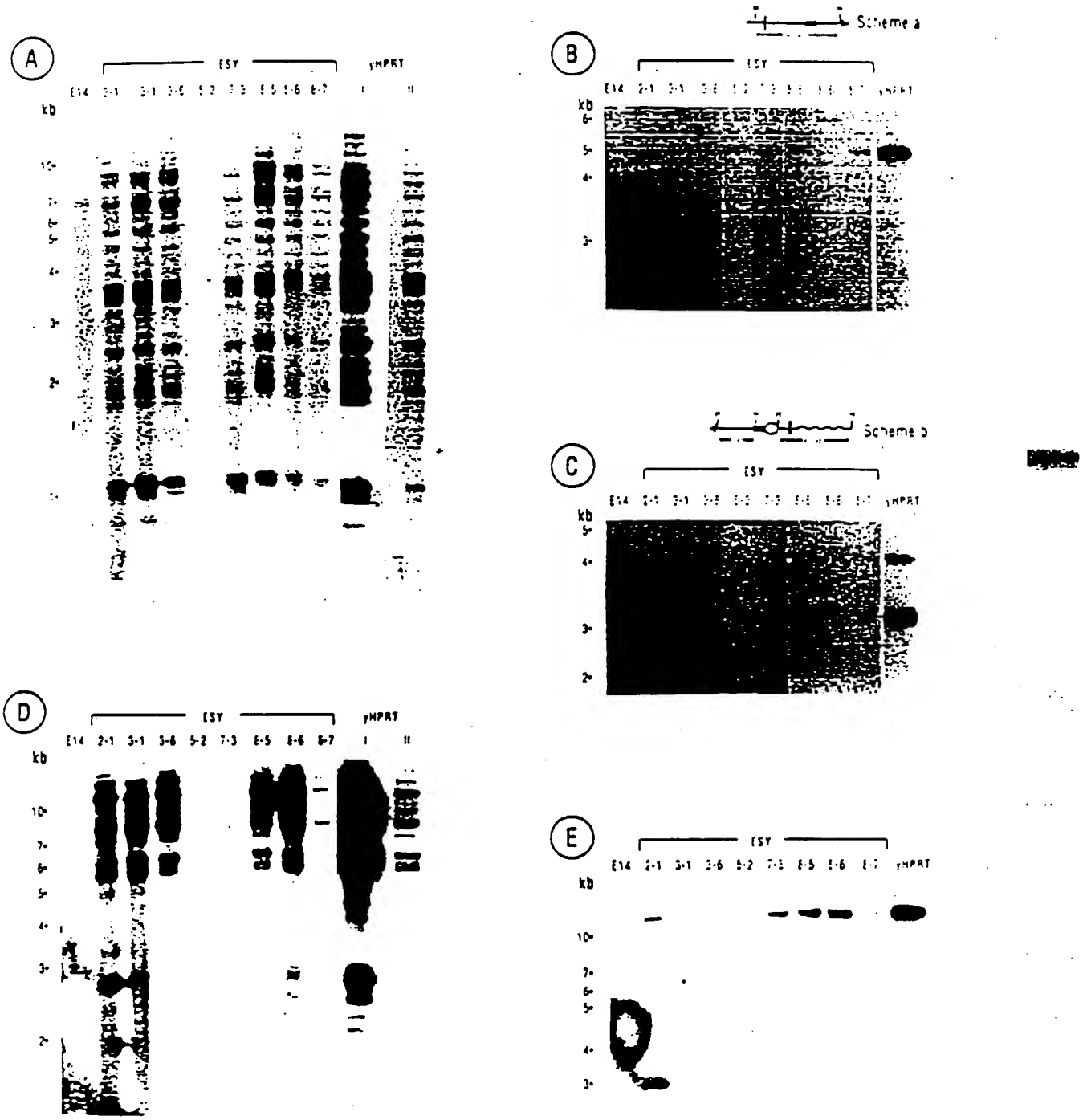

A
ESY
yHPRT
E14 2-1 3-1 3-6 5-2 7-3 8-5 8-6 8-7 I II
kb
B
Scheme a
ESY
E14 2-1 3-1 3-6 5-2 7-3 8-5 8-6 8-7 yHPRT
kb
C
Scheme b
ESY
E14 2-1 3-1 3-6 5-2 7-3 8-5 8-6 8-7 yHPRT
kb
D
ESY
yHPRT
E14 2-1 3-1 3-6 5-2 7-3 8-5 8-6 8-7 I II
kb
E
ESY
E14 2-1 3-1 3-6 5-2 7-3 8-5 8-6 8-7 yHPRT
kb

Figure 12

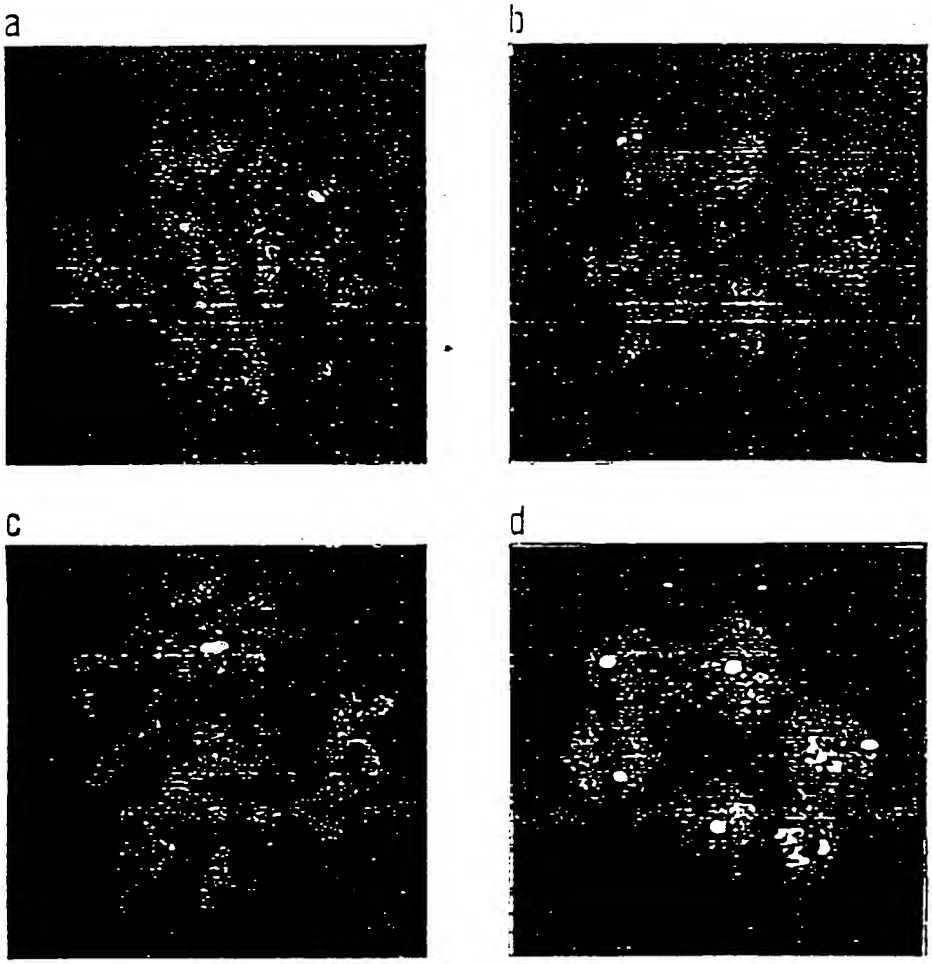
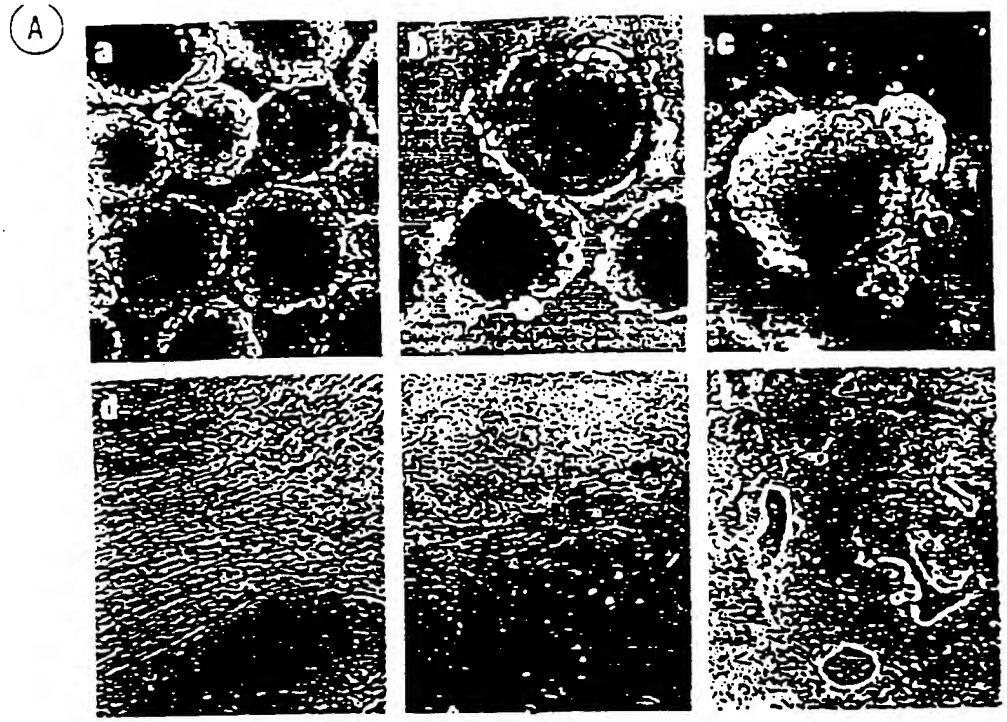
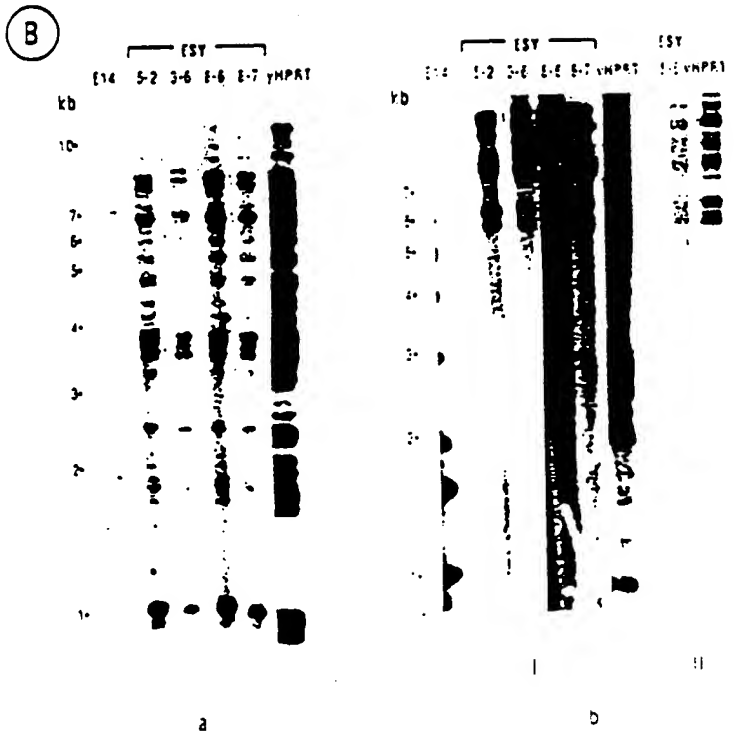



Figure 13



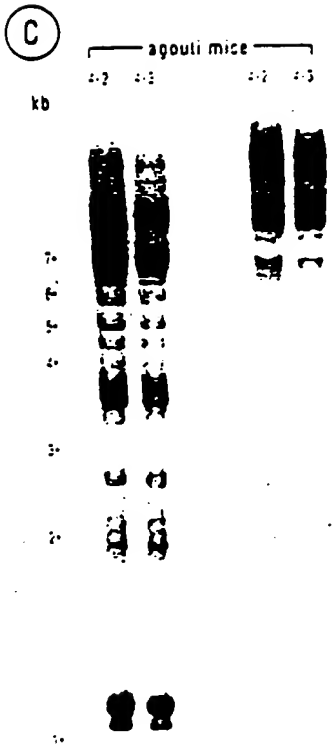




Figure 14

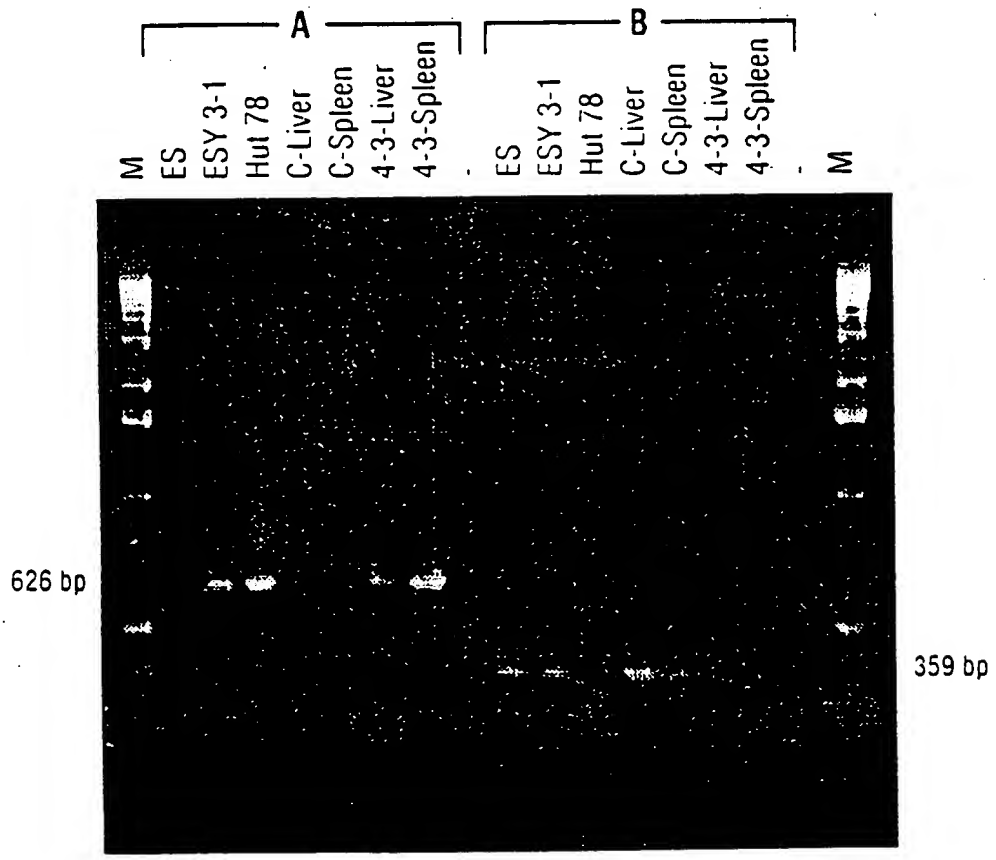
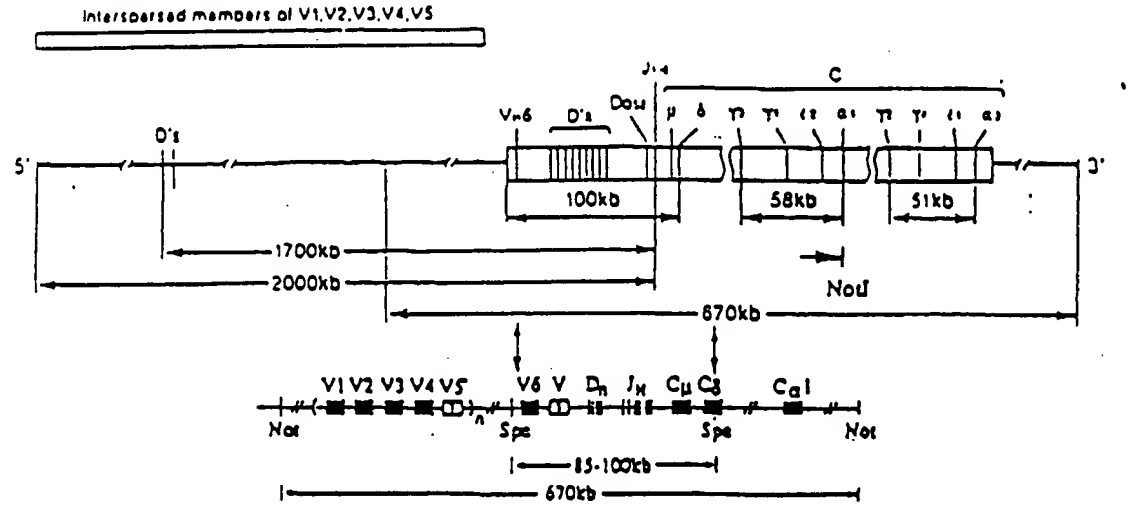
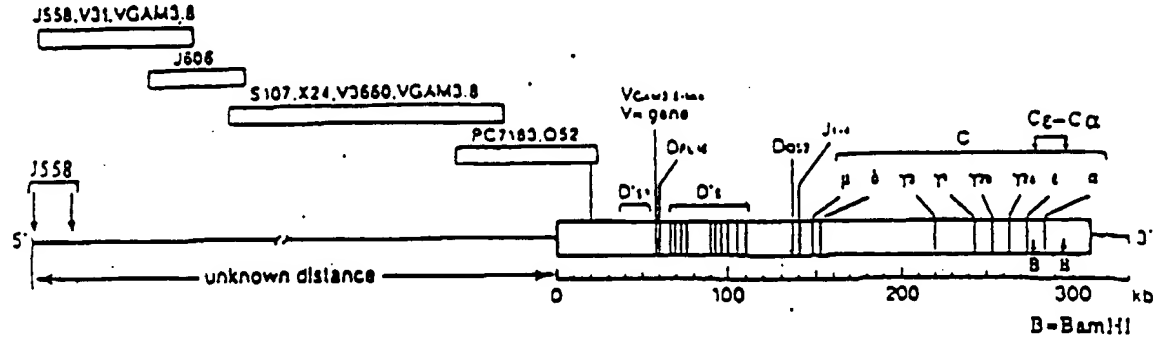

A
B
M
ES
ESY 3-1
Hut 78
C-Liver
C-Spleen
4-3-Liver
4-3-Spleen
ES
ESY 3-1
Hut 78
C-Liver
C-Spleen
4-3-Liver
4-3-Spleen
M
626 bp
359 bp

Figure 15

(A) Human heavy chain locus




(B) Mouse heavy chain locus




(C) Human heavy chain replacement YAC vector

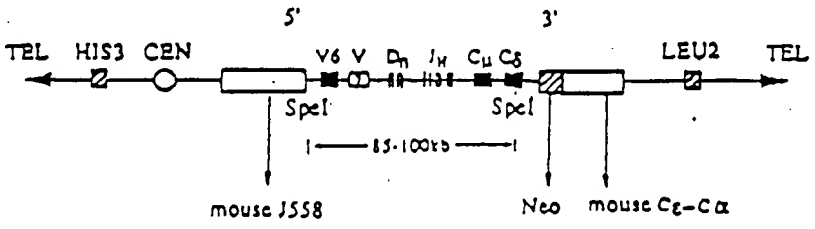



Figure 16

## Mouse Breeding Scheme

Cross IA.

heterozygous inactive Murine IgH
X
heterozygous inactive Murine IgK

MIgH (inactive) MIgK
MIgH MIgK
X
MIgH MIgK (inactive)
MIgH MIgK

↓

F1 (cross I A)

MIgH (inactive) MIgK (inactive)
MIgH MIgK

Cross I B.

heterozygous Human IgH
X
heterozygous Human IgK

MIgH MIgK HIgH
MIgH MIgK
X
MIgH MIgK HIgK
MIgH MIgK

↓

F1 (cross I B)

MIgH MIgK HIgH HIgK
MIgH MIgK

Cross II. F1 (cross I A) x F1 (cross I B)

↓

F2 Quadruple Heterozygotes

MIgH (inactive) MIgK (inactive) HIgH HIgK
MIgH MIgK

Cross III. Intercross F2 mice

↓

F3 DOUBLE Homozygotes

MIgH (inactive) MIgK (inactive) HIgH HIgK
MIgH (inactive) MIgK (inactive)

Figure 17

## MAMMALIAN HOST GENOTYPES

| | Hetero- or Hemi-zygous Mice | Intercross Product Mice* |
|---|---|---|
| I. | ΔmIgL mIgH<br>mIgL mIgH | ΔmIgL mIgH<br>ΔmIgL mIgH |
| II. | mIgL ΔmIgH<br>mIgL mIgH | mIgL ΔmIgH<br>mIgL ΔmIgH |
| III. | mIgL mIgH hIgH<br>mIgL mIgH | mIgL mIgH hIgH<br>mIgL mIgH hIgH |
| IV. | mIgL mIgH hIgL<br>mIgL mIgH | mIgL mIgH hIgL<br>mIgL mIgH hIgL |
| V. | Animal I X Animal II | |
| | ΔmIgL mIgH<br>mIgL ΔmIgH | ΔmIgL ΔmIgH<br>ΔmIgL ΔmIgH |
| VI. | Animal III X Animal V | |
| | mIgL mIgH hIgH<br>ΔmIgL ΔmIgh | ΔmIgL ΔmIgH hIgH and ΔmIgL ΔmIgH hIgH<br>ΔmIgL ΔmIgH hIgH ΔmIgL ΔmIgH |
| VII. | Animal IV X Animal V | |
| | mIgL mIgH hIgL<br>ΔmIgL ΔmIgH | ΔmIgL ΔmIgH hIgL and ΔmIgL ΔmIgH hIgL<br>ΔmIgL ΔmIgH hIgL ΔmIgL ΔmIgH |
| VIII. | Animal VI X Anmial VII | |
| | ΔmIgL ΔmIgH hIgL hIgH<br>ΔmIgL ΔmIgH | ΔmIgL ΔmIgH hIgL hIgH<br>ΔmIgL ΔmIgH hIgL hIgH |
| | mIgL mIgH hIgL hIgH<br>ΔmIgL ΔmIgH | ΔmIgL ΔmIgH hIgL hIgH and ΔmIgL ΔmIgH hIgL hIgH<br>ΔmIgL ΔmIgH hIgL hIgH ΔmIgL ΔmIgH |
| IX. | Animal III X Animal IV | |
| | mIgL mIgH hIgL hIgH<br>mIgL mIgH | mIgL mIgH hIgL hIgH<br>mIgL mIgH hIgL hIgH |
| X. | Animal II X Animal IX | |
| | mIgL ΔmIgH hIgL hIgH<br>mIgL mIgH | mIgL ΔmIgH hIgL hIgH and mIgL ΔmIgH hIgL hIgH<br>mIgL ΔmIgH hIgL hIgH mIgL ΔmIgH |
| XI. | Animal I X Animal IX | |
| | ΔmIgL mIgH hIgL hIgH<br>mIgL mIgH | ΔmIgL mIgH hIgL hIgH and ΔmIgL mIgH hIgL hIgH<br>ΔmIgL mIgH hIgL hIgH ΔmIgL mIgH |

*Not all possible genotypes from intercrosses are shown.

Δ = functionally inactive locus
m = mouse endogenous gene
h = human transgene
IgH = immunoglobulin heavy chain
IgL = immunoglobulin light chain

FIGURE 18